

PCTWORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification ⁷ : A61K 39/395, A61P 31/14	A1	(11) International Publication Number: WO 00/25816 (43) International Publication Date: 11 May 2000 (11.05.00)
(21) International Application Number: PCT/US99/25424 (22) International Filing Date: 29 October 1999 (29.10.99) (30) Priority Data: 60/106,266 30 October 1998 (30.10.98) US (71) Applicant: THE UNIVERSITY OF MIAMI [US/US]; Suite 906, 1400 N.W. 10th Avenue, Miami, FL 33101 (US). (72) Inventors: PINNA, Antonio; 7801 SW 51 Avenue, Miami, FL 33143 (US). RICORDI, Camillo; 3734 Matheson Avenue, Miami, FL 33133 (US). TZAKIS, Andreas, G.; 19 Samana Drive, Miami, FL 33133 (US). (74) Agents: MORDUCH, Ruth, N. et al.; Pillsbury Madison & Sutro LLP, 1100 New York Avenue, N.W., Washington, DC 20005 (US).		(81) Designated States: CA, JP, European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE). Published <i>With international search report. Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.</i>
(54) Title: TREATMENT OF HEPATITIS C BY ADMINISTRATION OF ANTI-IL-2 RECEPTOR MONOCLONAL ANTIBODY AND AN ANTIVIRAL COMPOUND (57) Abstract A novel method using anti-interleukin 2 receptor monoclonal antibodies and at least one antiviral compound for treating patients having severe viral hepatitis C is described. The inventive method may be used to treat recurrent hepatitis C following liver transplantation, thereby reducing complications in recipients of liver allografts.		

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania	ES	Spain	LS	Lesotho	SI	Slovenia
AM	Armenia	FI	Finland	LT	Lithuania	SK	Slovakia
AT	Austria	FR	France	LU	Luxembourg	SN	Senegal
AU	Australia	GA	Gabon	LV	Latvia	SZ	Swaziland
AZ	Azerbaijan	GB	United Kingdom	MC	Monaco	TD	Chad
BA	Bosnia and Herzegovina	GE	Georgia	MD	Republic of Moldova	TG	Togo
BB	Barbados	GH	Ghana	MG	Madagascar	TJ	Tajikistan
BE	Belgium	GN	Guinea	MK	The former Yugoslav Republic of Macedonia	TM	Turkmenistan
BF	Burkina Faso	GR	Greece	ML	Mali	TR	Turkey
BG	Bulgaria	HU	Hungary	MN	Mongolia	TT	Trinidad and Tobago
BJ	Benin	IE	Ireland	MR	Mauritania	UA	Ukraine
BR	Brazil	IL	Israel	MW	Malawi	UG	Uganda
BY	Belarus	IS	Iceland	MX	Mexico	US	United States of America
CA	Canada	IT	Italy	NE	Niger	UZ	Uzbekistan
CF	Central African Republic	JP	Japan	NL	Netherlands	VN	Viet Nam
CG	Congo	KE	Kenya	NO	Norway	YU	Yugoslavia
CH	Switzerland	KG	Kyrgyzstan	NZ	New Zealand	ZW	Zimbabwe
CI	Côte d'Ivoire	KP	Democratic People's Republic of Korea	PL	Poland		
CM	Cameroon	KR	Republic of Korea	PT	Portugal		
CN	China	KZ	Kazakhstan	RO	Romania		
CU	Cuba	LC	Saint Lucia	RU	Russian Federation		
CZ	Czech Republic	LI	Liechtenstein	SD	Sudan		
DE	Germany	LK	Sri Lanka	SE	Sweden		
DK	Denmark	LR	Liberia	SG	Singapore		
EE	Estonia						

TREATMENT OF HEPATITIS C BY ADMINISTRATION OF ANTI-IL-2 RECEPTOR MONOCLONAL ANTIBODY AND AN ANTIVIRAL COMPOUND

5

BACKGROUND OF THE INVENTION

1. Field of the Invention

The invention relates to a new use for existing monoclonal antibodies known to prevent or treat immunological rejection of transplanted organ and tissues. Specifically, the invention provides for the first time a method of using an anti-IL-2 receptor monoclonal antibody for reversing severe viral hepatitis C.

2. Description of Related Art

Recent advances in the field of organ transplantation have lead to the development of new therapy regimens effective in preventing acute rejection episodes in transplant recipients. Several preparations of antibodies are known for use as immunosuppressive agents in treating acute graft rejection. Some of these antibodies interact with certain lymphoid cells, thereby impairing cell function. For example, lymphocyte immune globulin (antithymocyte globulin) can sharply reduce the number of thymus-derived lymphocytes (T-cells) in treated patients and inhibit normal responses of such T-cells. Patients receiving a short course (induction) antithymocyte globulin are typically on other maintenance immunosuppressive regimens (6). The U.S. Food and Drug administration approved in 1998 a commercial form of an anti-interleukin-2 (IL-2) receptor monoclonal antibody for use in kidney transplantation protocols. This anti-IL-2 receptor monoclonal antibody binds to IL-2 receptors to prevent stimulation of T-lymphocyte production in response to a foreign organ graft. T-lymphocytes are major immunological players in the biological mechanisms responsible for organ transplant rejection. Anti-IL-2 receptor monoclonal antibodies, such as basiliximab (Simulect®, Novartis), may be added to current immunosuppressive regimens containing cyclosporine and prednisone for reducing acute rejection episodes

in renal transplant recipients. Such regimens appear effective in reducing irreparable damage to allografts subjected to acute rejection, improving post-transplant kidney function.

5 The addition of basiliximab to current immunosuppressive regimens permits organ recipients to be treated with a two-dose, four-day regimen in lieu of prior five-dose antibody regimens lasting 10 weeks. Basiliximab, cyclosporine and prednisone combination therapies reduce the incidence and severity of acute rejection episodes without increasing rates of infection and other associated consequences of nonspecific immunosuppression.

10 Similarly, clinical trials of a combination regimen containing daclizumab (Zenapax®, Hoffman-la Roche), mycophenolate mofetil, cyclosporine and corticosteroids have shown such a regimen to be efficacious in reducing acute rejection episodes within six months of kidney transplants. SMARTTM Anti-Tac, the humanized monoclonal antibody (daclizumab) active ingredient of Zenapax®, binds to the alpha
15 subunit of high affinity interleukin-2 receptors expressed on activated T-lymphocytes. Daclizumab antagonizes binding of IL-2 to high affinity IL-2 receptors, thereby suppressing T-lymphocyte activity against allografts.

Daclizumab-based therapy regimens improve organ transplant survival without unduly increasing therapy toxicity. Such humanized monoclonal antibodies have a
20 longer half-life and are less likely to be targeting by a patient's immune system than non-humanized antibodies, such as previously tested mouse monoclonal antibodies. Humanized immunoglobulins are disclosed in, for example, U.S. Patent Nos. 5,530,101 and 5,585,089.

Recurrence of hepatitis C following liver transplantation remains a feared
25 complication in recipients of liver allografts. Frequent recurrence of hepatitis C infection following orthotopic liver transplantation (OLT) is common, affecting up to 75% of transplant recipients (1). Hepatitis C viral RNA is detected in 95% to 100% of liver transplant patients suffering from recurrent hepatitis C. Traditionally, severe hepatitis C recurrence following liver transplantation is treated by reduction or
30 discontinuation of immunosuppressive therapy and a combination of antiviral drugs such as alpha interferon and ribovirin, a regimen which has proven to be ineffective.

Severe hepatitis C or cirrhosis secondary to hepatitis C continues to be a complicating factors in liver transplantation.

As noted herein, antiviral therapy for recurrent hepatitis C viral infection post-orthotopic liver transplantation has failed miserably to date. Cholestatic hepatitis has
5 an incidence of 8% to 13% with a high morbidity level. Characteristics indicative of cholestatic hepatitis include early hepatitis C virus recurrence, moderate to high bilirubin levels, renal insufficiency, ascites, and elevated serum hepatitis c viral RNA concentrations (3).

Recent experimental evidence suggests that viral specific cytotoxic T-
10 lymphocyte activation could be an important pathogenic mechanism of hepatocellular damage during chronic hepatitis C (2). Other studies indicate that only cells infected with non-cytopathic replicating viruses are destroyed by activated cytotoxic T-lymphocytes, due to viral induced expression of endogenous major histocompatibility complex (MHC) class I molecules (4). Cross-presentations of antigens expressed in
15 virally infected peripheral organs could explain activation of cytotoxic T-lymphocytes (5).

Therefore, a need exists for improved therapy for patients with hepatitis C and against hepatitis C virus recurrence in patients undergoing liver transplantations.

20 SUMMARY OF THE INVENTION

Treatment of patients with severe hepatitis C by an anti-IL-2 receptor antibody according to the invention may result in selective blockade of the effector cytopathic mechanism of hepatocellular damage, while concurrent anti-viral therapy eventually decreases the viral load in such patients.

25 An embodiment of the invention is a method of treating viral hepatitis C in a patient comprising combining an effective amount anti-IL-2 receptor monoclonal antibody with an effective amount of at least one antiviral compound for administration to a patient;

administering to a patient a dosage of the combination of anti-IL-2 receptor
30 monoclonal antibody and at least one antiviral compound; and

repeating administration of said dosage until liver function tests yield values in a clinically acceptable range,

wherein said dosage comprises administering either simultaneously or sequentially the anti-IL-2 receptor monoclonal antibody and the at least one antiviral compound.

Optionally, the method of treating viral hepatitis C in a patient may further comprise pretreating patients with an antihistamine or acetaminophen.

Another embodiment of the invention is a method of treating viral hepatitis C in a patient, wherein the patient is a liver transplant recipient suffering from recurrent cholestatic hepatitis C.

Yet another embodiment of the invention is a method of treating viral hepatitis C in a patient, wherein the effective amount of anti-IL-2 receptor monoclonal antibody comprises a dose of 2 mg /Kg of body weight.

Still another embodiment of the invention is a method of treating viral hepatitis C in a patient, wherein the dosage comprises a combination of an intravenously administered anti-IL-2 receptor antibody and at least one antiviral compound.

A further embodiment of the invention is a method of treating viral hepatitis C in a patient, wherein the dosage is repeated once weekly for up to six weeks.

A further embodiment of the invention is a method of treating viral hepatitis C in a patient, wherein a dosage comprises intravenously administering anti-IL-2 receptor monoclonal antibodies once every other week, and thereafter dosing is repeated monthly for at least one year.

DETAILED DESCRIPTION OF THE INVENTION

Recurrent cholestatic hepatitis C may be treated by reducing or suspending maintenance immunosuppression of patients, and administering anti-IL-2 receptor monoclonal antibody (Daclizumab) as a combination therapy with α -interferon and ribovirin. Doses of α -interferon typically are in the range of several million units. Ribovirin is administered at a level titrated to minimize side effects. Specifically, ribovirin may be administered in daily oral doses of 400 mg twice daily. Concurrently, for example, three million units of α -interferon may be injected subcutaneously three

times weekly. In addition, 2 mg/Kg of anti-IL-2 receptor monoclonal antibodies are administered intravenously, initially once per week, a frequency later reduced to once per month.

Patients may undergo an initial induction period, during which time one
5 injection of anti-IL-2 receptor monoclonal antibodies is given every other week for five weeks, and thereafter the injections are repeated monthly for at least one year. The treatment strategy may be individualized depending upon the viral load and biochemical response of each patient.

Other immunosuppressive drugs previously tested proved either to have no
10 effect on hepatitis C, or they undesirably increased or accelerated the progression of the disease. This generally required a decrease or discontinuation of immunosuppressive therapy.

Factors considered in the decision to use an anti-IL-2 receptor monoclonal antibody to block activated cytotoxic T cells against infected hepatocytes include: the
15 clinical conditions of the patients; the histology of a prior hepatic biopsy; and preliminary results of another study in which high dose donor bone marrow infusions significantly reduced the recurrence of hepatitis C. Selective blockade of activated T cells by the anti-IL-2 receptor monoclonal antibody daclizumab halted the progress of severe hepatic damage by hepatitis C, while allowing for other immune components to
20 maintain antiviral activity. This antibody therapy was clinically successful in patients, establishing the potential life-saving role of the inventive treatment in severe cases of hepatitis C.

Typically, the dosage of anti-IL-2-receptor antibody is similar or identical to dosages recommended by the FDA. Monitoring IL-2 receptor titer and the HCV-RNA
25 viral load can be helpful in titrating the dosage of anti-IL-2 receptor antibody and the length of treatment. The anti-IL-2-receptor antibodies are given to a patient by an intravenous route, either concurrently or sequentially, with the combined antiviral treatment comprising subcutaneous (s.c) administration of α -interferon and oral (po) administration riboviran. Preferred diluents or pharmaceutical carriers were those
30 suggested by the pharmaceutical manufacturer of the α -interferon. Preferably, patients are pretreated with an antihistamine or acetaminophen in order to alleviate the well-

known side effects of α -interferon. The treatment may initially comprise an injection of the anti-IL-2 receptor antibodies every other week for five weeks, and thereafter repeating dosing monthly for at least one year. This new use for anti-IL-2 receptor antibodies may be extended to patients with hepatitis C before a liver transplant becomes necessary, as well as to patients following liver transplantation to prevent recurrence of the disease.

Following confirmation of these pilot trials, anti-IL-2 monoclonal antibodies will be further tested in treating autoimmune diseases, including type 1 diabetes, multiple sclerosis, lupus, Crohn's disease, and the like. Heretofore, anti-IL-2 receptor antibodies have never been used for any of the above-described new applications. In patients with a primary autoimmune disease or with recurrent autoimmune disease after transplantation, a course of high dose intravenous anti-IL-2 receptor monoclonal antibody (2mg/Kg) may be instituted, to block the effector cells (cytotoxic T cells expressing the IL-2 receptor) until the clinical, biochemical and pathology expression of the autoimmune disorder disappear.

Blockade of the autoimmune response may enable preservation of the target cells, while therapeutic strategies are implemented to eliminate the underlying autoimmunity that could eventually be responsible for recurrence of the disease. Some of these strategies currently under investigation include bone marrow/stem cell transplantation as well as treatment with monoclonal antibodies, such as non depleting anti CD3 antibodies and/or antibodies directed at blockade of co-stimulatory pathways (e.g., anti CD154, anti-B7) or anti CD45RB. The invention is further illustrated by the following non-limiting examples.

EXAMPLES

Example I

Three male patients, ages 54, 54 and 53, underwent orthotopic liver transplantation for end stage liver disease secondary to hepatitis C.

Immunosuppression after transplant consisted of Tacrolimus (Prograf, Fujisawa) and glucocorticoids. All three patients developed recurrent hepatitis C within 60 days of receiving transplanted livers. Diagnosis of hepatitis C was made by liver biopsy,

elevated liver function test and determination of hepatitis C RNA viral load levels using a commercially available kit (Chiron Corporation, Emeryville, CA). Therapy was initiated in each patient at the time of recurrence comprising α -interferon at a dose of 3 million units s.c. three times a week + ribovirin 400 mg po twice a day. Concurrently, immunosuppression was decreased by reducing the administered doses of Tacrolimus to only 1 mg po a day and the steroidal therapy was stopped. None of the patients so treated responded to this therapy after an average of 1 month.

Additional liver biopsies confirmed recurrent hepatitis C, with moderate to severe activity and elevated hepatitis C RNA serum levels ($>120 \times 10^6$ Eq/ml, 31.7×10^6 Eq/ml and $>120 \times 10^6$ Eq/ml). Intravenous daclizumab at a dosage of 2 mg/Kg body weight once a week was started, while all remaining maintenance immunosuppression comprising Tacrolimus and steroids was ceased. Antiviral therapy remained unchanged. Daclizumab treatment continued weekly for five doses averaging 4 to 6 weeks in length until a decrease of the patient's bilirubin levels was observed, together with improvement of the histological markers of recurrent hepatitis C at the liver biopsy and clinical improvement of the patients. At that point, HCV-RNA viral load of the patients was significantly reduced or undetectable, despite being persistently high before the anti-IL2-receptor antibody treatment was started.

A remarkable clinical improvement was observed in all three patients between two and four weeks from the onset of daclizumab treatment. At the same time, only mild to minimal hepatitis was detected in follow-up liver biopsies. Significant decreases in direct bilirubin levels were measured in two of the three patients (from 30 to 10 mg/dL in one and from 5 to 0.5 mg/dL in the other) after three weeks of daclizumab therapy. Hepatitis C viral RNA levels decreased from $>120 \times 10^6$, 31.7×10^6 and $>120 \times 10^6$ Eq/ml to 2.2×10^6 Eq/ml, 7.6×10^6 Eq/ml and non-detectable levels, respectively. Two of the treated patients were discharged and are currently well at home, while the third was able to resume normal activity after two weeks of antibody treatment according to the invention.

Example II

The above-described daclizumab regimen was carried out on a total of 11 patients with recurrent cholestatic hepatitis C, each of whom had previously received α -interferon plus riboviran combination therapy. The mortality for recurrent cholestatic hepatitis C is typically about 90%. Of the 11 patients receiving the inventive therapy of 2 mg/kg body weight intravenous daclixumab once weekly for 4 to 6 weeks, in combination with antiviral agents, only one patient failed to survive. That patient three months after treatment with Zenapax due to causes unrelated to the treatment (aspiration pneumonia). Three patients underwent liver retransplantation for either chronic rejection (2 patients) or untreatable cholestatic hepatitis C (1 patient).

Table 1 shows the modification of the liver pathology after treatment, the evolution of the clinical picture of the 11 patients and the viral RNA HCV load obtained before and after the antiviral/anti-IL-2 receptor monoclonal antibody regimen.

Table 1

Bilirubin, HCV titer and liver biopsy before treatment with Zenapax				Bilirubin, HCV titer and liver biopsy after treatment with Zenapax			Outcome	
Name	Bil	HCV	Liver Biopsy	Bil	HCV	Liver Biopsy	Re-transplant	Status
Dea	25	120,000,000	Cholest.HCV	3	250,000		no	dead
Lam	20	100,000,000	Cholest.HCV	20	200,000	chr.rej.	yes	alive
Ass	10	120,000,000	Cholest.HCV	0.8	200,000	normal	no	alive
Gar	5	15,000,000	Cholest.HCV	0.8	600,000	normal	no	alive
Rob	5	120,000,000	Cholest.HCV	0.7	200,000	chr.hcv	no	alive
Sho	18	84,400,000	Cholest.HCV	1	5,000,000	chr.hcv	yes	alive
Edw	10.5	120,000,000	Cholest.HCV	0.5	49,000,000	normal	no	alive
Joh	14	110,000,000	Cholest.HCV	4	8,300,000	chr.hcv	no	alive
Smi	20	120,000,000	Cholest.HCV	20	3,700,000	chr.rej.	yes	alive
Gra	5	120,000,000	Cholest.HCV	1.3	26,800,000		no	alive
Ian	4	120,000,000	Cholest.HCV	1	8,300,000		no	alive

Use of the inventive method in these patients having severe recurrent hepatitis C after liver transplantation resulted in dramatic improvement in liver function tests

(bilirubin, AST, ALT), clinical conditions and renal function. Typically, hyperbilirubinemia occurs at blood levels of bilirubin greater than about 1.0 mg/dL. Severe hemolytic anemia usually does not produce sustained elevation of blood bilirubin levels greater than about 4.0 mg/dL, unless hepatic bilirubin clearance is compromised. However, patients experiencing liver dysfunction, such as those suffering from viral hepatitis in combination with a hemolytic disease, may have circulating levels of bilirubin approaching 100 mg/dL. The figures in Table 1 show the overall ability of the inventive therapy to reduce bilirubin levels in patients with hepatitis C to a clinically acceptable range. Prior to treatment, these patients were in critical condition, but resumed nearly normal lives following therapy according to the invention.

Thus, results indicate that anti-IL-2 monoclonal antibodies are effective for controlling cholestatic hepatitis C in patients following liver transplantation, as well as for controlling hepatitis C in patients before liver transplantation becomes necessary.

The foregoing description of the invention has been provided for the purposes of illustration. It is not intended to be exhaustive or to limit the invention to the precise embodiment(s) disclosed. The embodiment(s) were chosen and described in order to best explain the principles of the invention and its practical application, thereby enabling others skilled in the art to understand the invention for various embodiments and with various modifications as are suited to the particular use contemplated.

All journal articles, books, patents and publications cited herein are hereby incorporated in their entirety herein by reference.

References

1. Ascher NL, Lake, JR, Emond J, Roberts J. Liver Transplantation for Hepatitis C Virus-Related Cirrhosis. *Hepatology* 1994; 20: 245-275.
2. Asanza CG, Garcia-Monzon C, Clemente G, Salcedo M, Garcia-Buey L, Garcia-Iglesias C, Banares R, Alvarez E, Montero-otero R. Immunohistochemical Evidence of Immunopathogenic Mechanisms in Chronic Hepatitis C Recurrence After Liver Transplantation. *Hepatology* 1997; 26(3): 755-763.

3. Schluger LK, Sheiner PA, Thung SN, Lau JYN, Min A, Wolf DC, Fiel I, Zhang D, Gerber MA, Miller CM, Bodenheimer HC. Severe Recurrent Cholestatic Hepatitis C Following Orthotopic Liver Transplantation. *Hepatology* 1996; 23:971-976.
- 5 4. Zinkernagel RM, Doherty PC. The Discovery of MHC Restriction. *Immunology Today* 1997; 18:14-17.
5. Carbone RF, Kurts C, Bennett SRM, Miller J, Heath WR. Cross-Presentation: A General Mechanism for CTL Immunity and Tolerance. *Immunology Today* 1998; 19: 368-373.
- 10 6. Immunosuppressive Agents. In, *Goodman Gilman's The Pharmacological Basis of Therapeutics*. (Gilman A, Rall T, Nies A, and Taylor P, eds.) Pergamon Press, Inc. New York, 1990, pp. 1270-1276.

What is claimed is:

1. A method of treating viral hepatitis C in a patient comprising:
administering to a patient a dosage of an effective amount of an anti-IL-2 receptor monoclonal antibody and an effective amount of at least one antiviral compound; and
repeating administration of said dosage until liver function tests yield values in a clinically acceptable range,
wherein said dosage comprises administering either simultaneously or sequentially said anti-IL-2 receptor monoclonal antibody and said at least one antiviral compound.

2. The method according to claim 1, wherein said effective amount of anti-IL-2 receptor monoclonal antibody comprises a dose of 2 mg/Kg of body weight.
3. The method according to claim 1, wherein said dosage comprises intravenous administration of said anti-IL-2 receptor antibody.
4. The method according to claim 1, wherein said dosage comprises administration of said at least one antiviral compound via a route selected from the group consisting of oral administration and subcutaneous injection.
5. The method according to claim 1, wherein said dosage is repeated once weekly for up to six weeks.
6. The method according to claim 1, wherein a dosage comprises intravenously administering anti-IL-2 receptor monoclonal antibodies once every other week, and thereafter dosing is repeated monthly for at least one year.

7. The method according to claim 1, further comprising pretreating patients with an antihistamine or acetaminophen.
8. A method of treating recurrent cholestatic hepatitis C in a liver transplant recipient comprising:
 - administering to a liver transplant recipient a dosage of an effective amount of an anti-IL-2 receptor monoclonal antibody and an effective amount of at least one antiviral compound; and
 - repeating administration of said dosage until liver function tests yield values in a clinically acceptable range,
 - wherein said dosage comprises administering either simultaneously or sequentially said anti-IL-2 receptor monoclonal antibody and said at least one antiviral compound.

INTERNATIONAL SEARCH REPORT

International Application No. —

PCT/US 99/25424

A. CLASSIFICATION OF SUBJECT MATTER

IPC 7 A61K39/395 A61P31/14

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 C07K A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	<p>O. UTERMOHLEN ET AL.: "T lymphocyte-mediated antiviral immune responses in mice are diminished by treatment with monoclonal antibody directed against the interleukin-2 receptor."</p> <p>EUROPEAN JOURNAL OF IMMUNOLOGY, vol. 24, no. 12, December 1994 (1994-12), pages 3093-3099, XP000857878</p> <p>Weinheim, Germany</p> <p>the whole document</p> <p style="text-align: center;">--- -/--</p>	1-8



Further documents are listed in the continuation of box C.



Patent family members are listed in annex.

* Special categories of cited documents:

- "A" document defining the general state of the art which is not considered to be of particular relevance
- "E" earlier document but published on or after the international filing date
- "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- "O" document referring to an oral disclosure, use, exhibition or other means
- "P" document published prior to the international filing date but later than the priority date claimed

- "T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
- "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
- "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.
- "&" document member of the same patent family

Date of the actual completion of the international search

22 February 2000

Date of mailing of the international search report

29/02/2000

Name and mailing address of the ISA

European Patent Office, P.B. 5818 Patentlaan 2
NL - 2280 HV Rijswijk
Tel. (+31-70) 340-2040, Tx. 31 651 epo nl,
Fax: (+31-70) 340-3016

Authorized officer

No01j, F

INTERNATIONAL SEARCH REPORT

International Application No

PCT/US 99/25424

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	X. ZERVOS ET AL.: "Comparison of tacrolimus with microemulsion cyclosporine as primary immunosuppression in hepatitis C patients after liver transplantation." TRANSPLANTATION, vol. 65, no. 8, 27 April 1998 (1998-04-27), pages 1044-1046, XP000857875 Baltimore, MD, USA the whole document ----	1-8
A	EP 0 449 769 A (SANDOZ LTD. ET AL.) 2 October 1991 (1991-10-02) page 8, line 39 - line 41 -----	1-8

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US 99/ 25424

Box I Observations where certain claims were found unsearchable (Continuation of Item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:
Remark: Although claims 1-8 are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.
2. ☐ Claims Nos.:
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of Item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

Information on patent family members

Int'l Application No
PCT/US 99/25424

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
EP 449769 A	02-10-1991	AT 98655 T	15-01-1994
		AU 635401 B	18-03-1993
		AU 7290991 A	19-09-1991
		CA 2038279 A,C	17-09-1991
		CY 1977 A	05-09-1997
		DE 69100768 D	27-01-1994
		DE 69100768 T	11-05-1994
		DK 449769 T	21-02-1994
		ES 2061216 T	01-12-1994
		FI 911275 A	17-09-1991
		FI 981799 A	21-08-1998
		HK 53797 A	02-05-1997
		HU 60768 A	28-10-1992
		HU 9500213 A	28-12-1995
		IE 65062 B	04-10-1995
		JP 2585475 B	26-02-1997
		JP 4316600 A	06-11-1992
		LU 90383 A	07-06-1999
		NZ 237434 A	26-08-1992
		PL 170321 B	29-11-1996
		PT 97034 A,B	31-10-1991
		ZA 9101947 A	25-11-1992